SEMIQUALITATIVE COLLECTION TECHNIQUES FOR BENTHIC MACROINVERTEBRATES: USES FOR WATER POLLUTION ASSESSMENT IN NORTH CAROLINA

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Abstract

Semiqualitative collection methods for benthic macroinvertebrates have been used in many types of water pollution studies in North Carolina. These techniques emphasize multihabitat collections and the use of both coarse and fine-meshed samples. These techniques have proven to be more cost-effective and have generated more useful data than conventional collection techniques. Examples of how data are used in several monitoring programs are presented and include trend monitoring, point source surveys (including toxicity reduction) and use attainability.

Introduction

Benthic macroinvertebrates have been collected using a variety of quantitative and qualitative techniques by many state and federal agencies. Quantitative approaches (i.e. Hester Dendy multiplates or Surber samples) are thought to be more precise and more amenable to statistical analysis; however, quantitative techniques are both time and cost intensive. Quantitative sampling is also usually habitat specific, resulting in a large portion of the aquatic community not being sampled. For example, Allan (1975) found that twelve Surber samples underestimated total taxa richness (from a variety of collection methods) by about 32%.

The North Carolina Division of Environmental Management (DEM) uses aquatic macroinvertebrates to assess water quality in streams and rivers. A large number of sites are sampled each year (300+) and the information gathered is used to document both spatial and temporal changes in water quality. To deal with this large volume of work, a new semiqualitative collection technique was developed. This technique samples most microhabitats using both coarse-mesh and fine-mesh samplers. Abundance values are qualitative (rare, common, abundant), but taxa richness information is quantitative.

<u>North Carolina Monitoring Programs</u>

The North Carolina Division of Environmental Management (DEM) conducts several types of water quality assessment programs. Emphasis has been directed towards measuring the effects of point source dischargers (and toxicity reduction) and trend (or ambient) monitoring. Water quality surveys are also conducted to assess non-point source pollution, water use attainment and spill assessment. The use of semiqualitative collection techniques for benthic macroinvertebrates has allowed biological monitoring data to be incorporated into each of these monitoring programs.

<u>Semiqualitative Method Description</u>

The method description presented here is an excerpt from a paper (in preparation) written by David Lenat. This paper provides a complete discussion of North

Carolina's qualitative collection technique.

This technique is intended for use only in wadable, freshwater streams. Sampling is easiest during periods of dry weather, as high flow conditions may severely impair sampling efficiency by making critical habitats inaccessible. Collections are designed to have a fixed number of samples at each site (10), although many of these are composite samples. Six different collection methods are utilized to collect qualitative samples from a variety of microhabitats. All samples are picked in the field.

Kick nets: Two kick samples are taken, usually in riffle areas. In very small streams, or in sandy areas without riffles, kicks are taken from root masses, "snags", or bank areas. All types of benthic macroinvertebrates can be collected with this sampling technique, but emphasis is placed on the collection of Ephemeroptera, Plecoptera and Trichoptera. Kick net samples are washed down in a large bucket sieve prior to being processed.

Sweep nets: Three samples are taken by physically disrupting an area and then vigorously sweeping through the disturbed area with a long-handled triangular net (approximately 1 mm mesh size). Sweeps are usually taken from bank areas and/or macrophyte beds. Bank areas are particularly important for the collection of species which prefer low current environments. In particular, samples are inspected for Chironomini, Oligochaeta, Odonata, mobile cased Trichoptera, Hemiptera, Sialis, Crustacea, and Ephemeroptera. Large rocks or bedrock areas with attached macrophytes (especially river weed, Podostemum) also may be sampled with the sweep net to look for Hydropsychidae, Baetidae and Ephemerellidae. A sweep net can also be used to collect gravel, which is inspected for stone-cased Trichoptera. The latter technique is used principally in the sandhills area of North Carolina, an ecoregion with a diverse trichopteran assemblage.

Fine-mesh sampler: Smaller organisms, especially Chironomidae, are sampled with a finer mesh (300 microns) and are field-preserved to increase picking efficiency. Rocks or logs with visible growths of periphyton, Podostemum or moss are washed into a large plastic basin (or bucket) partially filled with water, and the substrate is vigorously brushed or rubbed to dislodge all attached fauna. Any large particulate material (leaves, etc.) is washed and discarded. A single composite sample is made from several rocks and logs. The material remaining in the basin is poured through the fine mesh sampler, which is constructed of 4 inch PVC, and the water drained completely. The residue is quickly preserved in 95% alcohol by placing the PVC cylinder into another slightly larger container half filled with 95% alcohol. The sample soaks in alcohol for about five minutes, and then is backwashed with stream water into a picking tray. This method of field preservation requires only a small amount of reusable alcohol.

Sand samples: Sand habitats often contain a very distinct fauna, but extraction of this fauna, using dredges, cores, etc., can be very tedious. Sand substrates are sampled with a large bag constructed of fine mesh (300 microns) nitex netting. It can be quickly constructed from a one meter square piece of netting,

Table 1. Taxa richness criteria (10 samples) for assigning water quality classification to free-flowing, wadable North Carolina streams and rivers, July to September.

Class	EPT* Taxa Richness Mountain	EPT* Taxa Richness Piedmont	EPT* Taxa Richness Coastal	Total Taxa Richness Mountain	Total Taxa Richness Piedmont	Total Taxa Richness Coastal
Excellent	>41	>31	>27	>91	>91	>83
Good	32-41	24-31	21-27	77-91	77-91	68-83
Good-Fair	22-31	16-23	14-20	61-76	61-76	52-67
Fair	12-21	8-15	7-13	46-60	46-60	35-51
Poor	0-11	0-7	0-6	0-45	0-45	0-35

* Most intolerant groups = Ephemeroptera + Plecoptera + Trichoptera

folded in half and sewn together on the opposite side and the bottom. This bag is used like a Surber sampler, but the lack of a rigid frame allows for easy storage when folded. The bag is held (open) near the substrate, and the sand just upstream is vigorously disturbed. A composite sample is collected, utilizing 2 to 3 locations. The material collected (a lot of sand and a few organisms) is emptied into a large plastic container half-filled with water. A "stir and pour" elutriation technique is used in conjunction with the fine mesh sampler described above. After field preservation, the elutriate is checked for Chironomidae (especially <u>Rheosmittia</u>, <u>Harnischia</u> group and <u>Polypedilum</u> spp.), Oligochaeta, Gomphidae and some Ephemeroptera.

<u>Leaf-pack samples</u>: A large, coarse-meshed bucket sieve is used to wash down "aged" (decomposing) leaf-packs, sticks and small logs. Such samples are particularly helpful in large sandy rivers where many of the species are confined to "snags" (Benke et al. 1984, Neuswanger et al. 1982). This technique is good for finding "shredders", especially Tipulidae, Plecoptera and Trichoptera.

<u>Visual search</u>: Large rocks and logs are visually inspected for any associated invertebrates. Certain tightly adhering organisms may be collected only by this technique (Lepidoptera, Blephariceridae, <u>Leucotrichia</u>, <u>Psychomyia</u>). Decaying logs are picked apart, especially logs with loose bark. Freshwater sponges are inspected for Chironomidae (<u>Xenochironomus</u>), Trichoptera (<u>Ceraclea</u>) and Neuroptera. Rocks near the shore (in negligible current) will harbor certain Ephemeroptera and Odonata, and leaves near the shore may be primary habitat for some Gastropoda. In addition, a mussel search is conducted by careful visual inspection of the bottom.

<u>Sample processing/identification</u>: A standardized qualitative

collection consists of ten samples: two kicks, three sweeps, three fine mesh samples (two rock-log samples and one sand), one leaf-pack and visuals (considered as one sample). All samples are field picked with jeweler's forceps from white plastic or enamel trays and preserved in 6 dram vials. Organisms are picked in proportion to their abundance, but no attempt is made to remove all organisms. If an organism can be reliably identified as a single taxon in the field, no more than 10 individuals need to be collected.

Samples are identified in the lab, using species level identification when possible. Chironomidae and Oligochaeta are sorted under a dissecting microscope and representative individuals are slide mounted. During sample processing and identification, it is fairly simple to add some measures of abundance. As invertebrates are identified, they are recorded as abundant (>9), common (3-9), or rare (<3). Field notes can also be used to label exceptionally abundant (dominant) species. Total taxa richness and taxa richness for Ephemeroptera, Plecoptera and Trichoptera (EPT) are calculated and used to assign a biological classification to each station (see criteria development).

EPT surveys: A general assessment of water quality can also be obtained quickly using an abreviated method which involves collecting only four samples (1 kick, 1 sweep, 1 leaf-pack and visuals) and identifying only Ephemeroptera, Plecoptera and Trichoptera. This technique, called an EPT survey, is used to supplement full qualitative surveys at a greater number of locations. The information can be used to assign relative water quality classifications and determine if additional or supplemental surveys should be conducted. In some instances the EPT survey may be used to assess impacts to the intolerant EPT groups. At this point, only preliminary comparisons have been made between EPT values collected during full qualitative (10 samples) and EPT surveys (4 samples). These comparisons (N=10) suggest that full surveys collect 1.1 to 1.3 times more EPT taxa. More testing in specific ecoregions and stream orders need to be conducted.

Criteria Development

Criteria have been developed for three major geographic regions (NC DEM 1986, Table 1) to relate both total taxa richness and EPT taxa richness to five water quality classifications: Excellent, Good, Good-Fair, Fair and Poor. These criteria have been used since 1983 with little modification. Upper (Excellent classification) and lower (Poor classification) limits were established based on collections in each region at both unstressed and highly polluted sites. The other three classes were then defined by dividing the remaining taxa richness ranges into three equal groupings.

Data collected from unimpacted sites suggest that there is a relationship between region and the taxa richness of the benthic macroinvertebrate community. This assumption was tested by comparing average taxa richness data from summer collections (July to September) at unstressed sites in the mountains (7 sites), the piedmont (6 sites) and the inner coastal plain (4 sites). Areas that had intermediate characteristics (upper piedmont) were not included in this analysis. The majority of the sites had been sampled in more than one year (up to six years) and average taxa richness values were utilized. "Unstressed sites" were defined as areas with no known chemical/physical alterations, having a high diversity of invertebrates and/or fish. Going east from the mountains to the coastal plain, taxa richness decreased for Ephemeroptera (20.6 , 15.8 and 10.1, respectively) and increased for Odonata (4.6, 8.2 and 11.1, respectively). Plecoptera, Trichoptera and Diptera were most diverse in the mountains, while Coleoptera, Crustacea and Mollusca were more diverse in the piedmont/coastal plain areas. The EPT subtotal was clearly higher in the mountains (44.7 as compared to 31.9 in the piedmont and 28.7 in the coastal plain), but total taxa richness differences were not as great (mountain = 98.9, piedmont = 93.3 and coastal plain = 91.0).

Water Quality Assessments Using Benthic Macroinvertebrates

Semiqualitative collection techniques have been used successfully in a number of monitoring programs. The most ambitious program in North Carolina is the Benthic Macroinvertebrate Ambient Network (BMAN). However, these collection methods are flexible enough to allow for the use of benthic macroinvertebrate data in a number of other programs. Several of these include point source monitoring (including toxicity reduction), use attainability analyses and water use classification.

Benthic Macroinvertebrate Ambient Network (BMAN)

The North Carolina Division of Environmental Management (DEM) maintains 350 ambient locations in seventeen (17) major river basins. Water quality data is collected from each site by personnel in 7 regional offices on a monthly or quarterly basis. Benthic macroinvertebrate samples have been collected from a total of 208 of these locations to assess long term trends (since 1982) in water quality. Samples are collected from 80-100 of these sites each year. These locations are staggered each year (collections are made either on an annual, biennial or triennial basis so that greater state wide coverage is feasible). BMAN samples are collected during summer months to lessen temporal variation between years and document worst-case (low flow, high temperature) conditions. Between-year changes in the composition of the benthic community structure, including differences in total taxa richness (ST and SEPT, and subsequent bioclassification) as well as the occurrence of dominant taxa or "indicator groups" are used in the analyses. Between-year analyses also include important flow related variables. These include changes in current velocity, changes in non-point contributions and changes in the length of recovery zones below point sources.

Biological data are tabulated each year summarizing trends in water quality. In 1986 for example, positive trends were noted at eighteen (18) BMAN locations while negative between year trends were noted at only four (4) stations. These data were collected during an extremely low flow period in North Carolina, which probably tended to reduce the effects of non-point sources of pollution. The effects of point sources also may have been increased at some sites by reducing effluent dilution. Additionally,

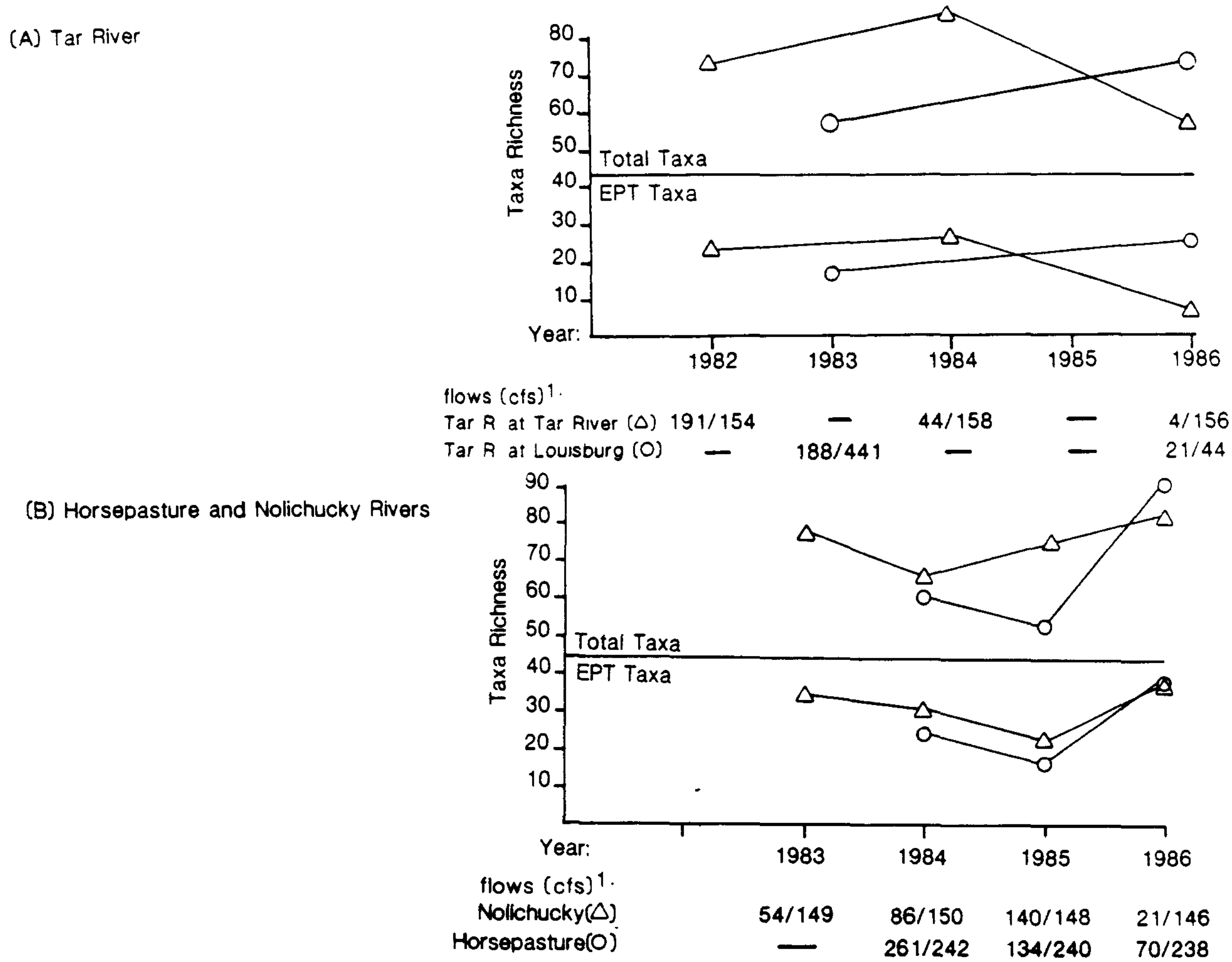
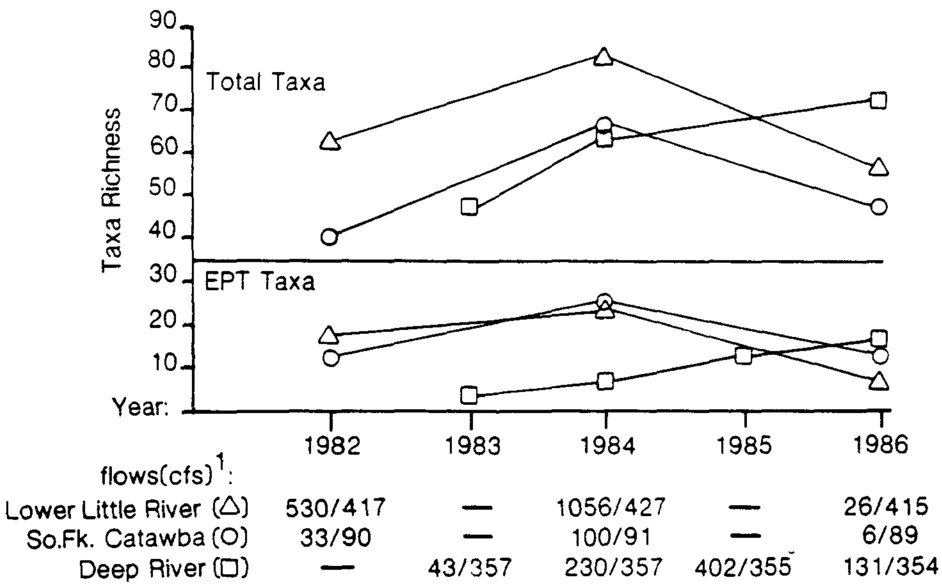


Fig. 1. Trend analysis effects of flow and non-point source impact on benthic taxa richness.

between-year changes in benthic fauna are compared to changes in chemical water quality data.

Biological data have been useful in documenting between-year changes in water quality, although interpretation can not be limited to biological data. For illustration, several examples are provided. Figure 1A and 1B illustrate taxa richness data and flow at several stations from watersheds having significant non point source (sedimentation) contributions. Data from 1986 indicate that reduced flows significantly reduced taxa richness from the Tar River at Tar River site (Fig. 1A) because flows were almost negligible (3% of the average flow), thus reducing the number of taxa dependant on flow. However, taxa richness values were greater at the downstream Tar River at Louisburg site in 1986 than in 1983. Flow, although reduced at Louisburg in 1986, was sufficient enough to maintain flow-dependant taxa (i.e. filter-feeders), but reduced the effects of non point source pollutants. Low flow conditions improved taxa richness and, in this case, increased the bioclassification from Good-Fair to Good.



1 Flows are expressed as f30/average flow = average flow 30 days prior to benthos collection/average flow for period of record.

Fig. 2. Trend analysis effects of flow and point source impacts to benthic taxa richness Lower Little, South Fork Catawba and Deep Rivers.

Data are illustrated from two mountain rivers (the Horsepasture and Nolichucky Rivers) in western North Carolina in Figure 1B. These data indicate that during high flow years (1984 and 1985) non-point source contributions are greater, thus reducing taxa richness values. However, taxa richness values increased in 1986 during extremely low flow conditions. Much of the Horsepasture River watershed is currently being developed for second homes and tourism and therefore subject to impacts from sedimentation. Continued monitoring should detect the effect of these activities on the instream fauna.

Figure 2 illustrates data from stations selected below major municipal dischargers. In 1984, during high flow, the benefits of dilution to instream fauna are evidenced at both the Lower Little and South Fork Catawba River locations. Instream waste concentrations were greater during low flow years (1982 and 1986) and taxa richness values were lower. The exception to these trends are data from the Deep River. Facility upgrades have improved water quality in the basin (NRCD 1988a), resulting in increased taxa richness and improved water quality even during low flow periods with less dilution of impacts.

In 1984, facility upgrades (denitrification) were also completed on the major municipal above the South Fork Catawba River station. Lower ammonia levels and greater flow (dilution) in 1984 resulted in increased taxa richness. Also, many toxic tolerant chironomids (Cricotopus bicinctus,

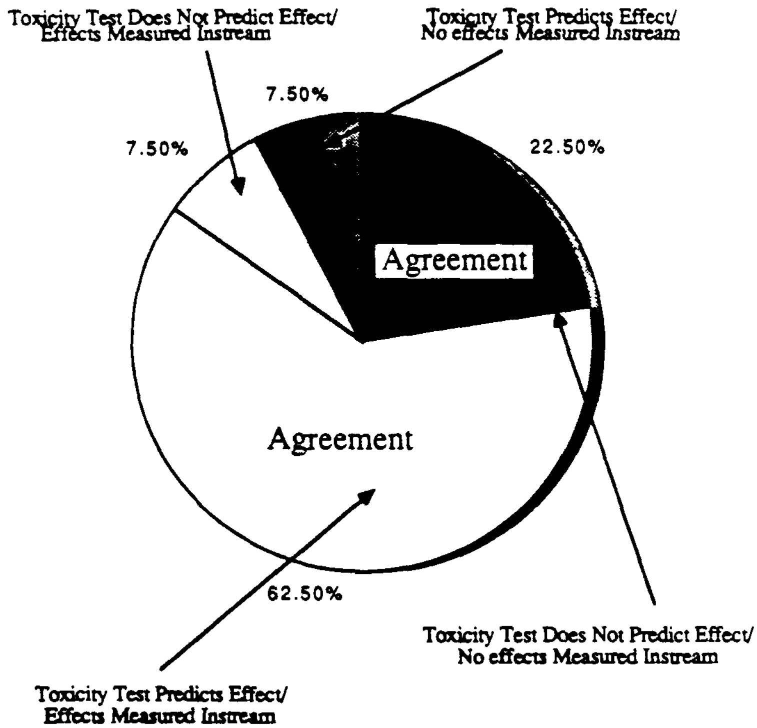


Fig. 3. Whole effluent toxicity measurements and resultant instream impacts as reflected by benthic macroinvertebrate populations.

C. tremulus, C. varipes and Polypedilum illinoense) were abundant in 1982, but absent or reduced in abundance in 1984. However, taxa richness was again reduced in 1986 during low flow and the toxic tolerant chironomids were replaced by enrichment indicators (Chrionomus sp., Tribelos sp., Tanytarsus sp. and Limnodrilus hoffmeisteri). These latter observations indicate the value of using "indicator groups" in trend analysis.

Point Source Monitoring Programs

The primary responsibility of many state biological monitoring programs is to assess point source impacts. Simply assessing the effects of a point source using the indigenous aquatic fauna can be achieved using several techniques. However, qualitative collection techniques have allowed biological data in North Carolina to be used effectively to direct and/or supplement management strategies. For example, benthic macroinvertebrate data is used as a screening tool to occasionally direct more labor intensive chronic toxicity studies, to supplement effluent toxicity testing, to assess the effectiveness of facility upgrades and also to supplement water quality investigations of fish kills or other episodic events detrimental to water quality.

Currently the use of instream benthic surveys to test for toxicity are not required in the NPDES permitting process; however, benthic data has been collected to supplement whole effluent toxicity testing at 40 North Carolina facilities. This information is used to identify impacts not addressed by numerical standards. Figure 3 illustrates strong agreement (85%) between results of chronic toxicity testing using Ceriodaphnia which predict instream toxicity and benthic macroinvertebrate surveys which have measured instream toxicity (Eagleson et al. 1988). Only in six of the 40 surveys (15%) did effluent tests and benthos surveys not agree. In two of the three instances where benthos surveys did not detect instream toxicity predicted by assay, poor upstream water quality was noted which masked any downstream toxicity.

One goal of the 1972 amendments to the Federal Water Pollution Control Act was to require secondary effluent limits for all wastewater treatment plants. However, this requirement resulted in a great deal of debate over whether or not meeting secondary effluent limits (at a considerable expense to municipalities) would result in better water quality. To test the effectiveness of these additional controls, seven biological surveys were conducted (and used to supplement chemical surveys) before and after facility upgrades (NRCD 1984). The results of these investigations indicated that moderate to substantial instream improvements were observed at six of the seven facilities. Several more recent investigations also have noted instream improvements following facility modifications and compliance. The use of benthic macroinvertebrate surveys to test for instream improvements following facility modification has proven to be an efficient, cost-effective monitoring tool.

Benthic macroinvertebrate surveys are also used to assess, and identify causes, of fish kills or spill events. If proper protocols are taken, benthic information can be collected and processed rapidly, resulting in the information getting to the enforcement agency often times quicker than results of chemical or fish tissue surveys.

Use Attainability and Water Use Classification

The North Carolina Division of Environmental Management (DEM) has the responsibility of determining water use classifications for all North Carolina surface waters. These uses include water supply, fishing (trout and non-trout), shellfish waters, water contact sports and Outstanding Resource Waters. DEM has the responsibility to assess water use attainment (i.e. are the uses being supported?) and to assess any proposed reclassifications. For example, recent regulations promulgated by EPA (November 1983) require a "use attainability analysis" to be conducted when uses are removed from a stream classification. Use attainability and water use classification involve a comprehensive analysis of physical, chemical and biological factors affecting the attainment of a use. Benthic macroinvertebrate data has played a key role in these analyses and biologists have been principle authors of reports recommending appropriate classifications by evaluating attainable use.

The most recent Water Quality Progress report in North Carolina (NRCD 1988b) indicated that 60.7% of the almost 37,000 miles of freshwater streams and rivers support their intended uses, 24.8% partially support, 4.7% do not

Table 2. Comparison of biology (mostly benthic macroinvertebrate data) vs. chemical data for freshwater stream water use evaluations (NRCD 1988b).

River Basin	Final Evaluation Based on Chemistry	Final Evaluation Based on Biology	Both Biology & Chemistry Data Available: Agree	Disagree: Biology with Lower Rating	Disagree: Biology with Higher Rating
Mountains					
Little Tennessee	16%	84%	70%	0%	30%
French Broad	48%	52%	17%	19%	64%
New	0%	100%	47%	0%	53%
Piedmont					
Broad	28%	72%	88%	0%	12%
Yadkin-Pee Dee	52%	48%	40%	2%	58%
Neuse	42%	58%	58%	0%	42%
Coastal					
Chowan	60%	40%	16%	0%	84%
Lumber	45%	55%	0%	27%	73%
Roanoke	41%	59%	29%	0%	71%
Overall	41%	59%	41%	4%	55%

support and 9.9% were not evaluated. Much of this information was based on biological data and, in particular, taxa richness data from several of the monitoring programs noted earlier. A comparison of how biological and chemical data were used to support uses in several watersheds is outlined in Table 2. Biological data, mostly benthic macroinvertebrates, were used nearly 60% of the time to support the intended use designations. In instances when biology and chemistry disagreed as to a particular use category, 55% of the time biological data were used to assign a higher use support category.

A final water use classification determination in which benthic macroinvertebrate data are used is the designation of Outstanding Resource Waters (ORW). North Carolina's Administrative Code (1986) states that the Environmental Management Commission may classify certain unique and special surface waters of the State as Outstanding Resource Waters (ORW) upon finding that such waters are of exceptional state or national recreational or ecological significance and that the waters have exceptional water quality. This new regulation gives benthic biologists in North Carolina the opportunity to collect samples from unique habitat, to collect rare or unusual taxa and use these and other data sources (including fisheries and fisheries habitat) to recommend appropriate classification.

Special Studies

The use of semiqualitative collection methods to process benthic samples rapidly has allowed our biological monitoring group to assist other federal or state water pollution agencies. Cooperative studies have been conducted with the Forest Service (effects of gypsy moth eradication methods to non-target aquatic insects), the Soil Conservation Service (watershed protection programs and non-point source implementation), the U.S. Geological Survey (acid rain effects on water quality and establishment of a pristine streams network) and local and state councils of governments.

Summary

North Carolina's semiqualitative collection technique for benthic macroinvertebrates was developed to provide a rapid, but reliable assessment of water quality. This technique has proven to be flexible enough to allow biological monitoring data to be used in a number of monitoring programs. These programs include a benthic macroinvertebrate ambient monitoring network which annually summarizes trends in water quality. Ambient data have noted temporal variation in water quality, including flow related variables such as non-point source contributions. Benthic macroinvertebrate data are also used to supplement toxicity testing by measuring instream effects. Effluent toxicity and instream benthic data have an 85% agreement rate and are used by managers to identify impacts not addressed by numerical standards. Semiqualitative collection methods for benthic macroinvertebrates are also used to determine appropriate use classification in use attainability studies in addition to assessing instream improvements due to facility upgrades.

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